

Z/I Imaging PhotoScan 2001 scanner instructions (5 pages total)

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All first-time users should arrange for training from a qualified user from their working group to receive training prior to scanning. AFTER users have been trained and are competent using the scanner, they will be given access to Lilly B-216.

Bulbs that come with scanner are different than older scanner!

Philips DDL 20V150W-3 (Made in Japan)

When a bulb fails, enter in the logbook and ask Paul to change the bulb.

Do's:

- Clean up after yourself.
- Use gloves at all times when working inside the scanner/handling negatives.
- Enter your session into the log book.
- Sign up for no more than 1 session at a time.
- Cancel your reservations promptly if you can't use your reservation.
- Report any problems you find with the scanner to Paul and enter them into the logbook.

Don'ts:

- No eating/drinking near scanner.
- Never turn off the upper (tan) computer that drives the scanner.
- Do not improvise – if you can't figure it out, get help.
- Do not install any software or download files onto the PC.
- Do not add things to the “desktop” of the computer.
- Do not write into the system disk (C:) or the scratch disk (D:). Your files should be written in E: and F: instead (see scanning instructions).
- **Do not read e-mail, play games, listen to CDs with the PC, etc.**
- The scanner is extremely sensitive to the configuration of the PC driving it. WE HAVE BEEN WARNED BY THE COMPANY NOT TO MODIFY THE CONFIGURATION OF THE PC. THEY WILL NOT SERVICE THE INSTRUMENT IF IT HAS BEEN MODIFIED IN A MANNER INCONSISTENT WITH THE SCANNER REQUIREMENTS. Ask an experienced scanner user before exploring all the new features of the software. DO NOT change the configuration without notice.

Violation of these rules may result in loss of scanner privileges.

Before scanning:

- 1) Confirm that the scanner is not currently in use.
The scanner should be turned off after every scanning session, unless the next person scanning has showed up before the last person finishes.
- 2) Enter the date, your name, your supervisor's name into the log.
(Complete log entry after scanning – including number of micrographs scanned and scanner performance.)

How to turn on the scanner:

- 1) Turn scanner on first (before turning on PC) – green rocker switch on left side of the scanner.
- 2) **After** scanner has run self-diagnostics (indicated on top of scanner at front left), turn on the PC (upper rocker switch behind black door).
The PC boots up with Windows 2000.

How to log on to the PC:

ctrl-alt-delete (this brings up the log in prompt)

Username: ZI User

Password: get from Paul

Make sure the PC boots up properly before loading films, etc. If it doesn't, enter the symptoms in the log and notify Paul.

How to load films:

- 1) Prepare scanner and negative for scanning (WEAR GLOVES!):
 - A solution 100% ethanol or glass cleaner (such as Windex) may be used to clean the glass plates. Be extremely careful not to introduce scratches onto the plates – they are very expensive, optical-quality glass. Any user who finds any scratch on the surface of the glass should write this down in the log book.
 - Kimwipes or other high lint paper is not good for cleaning the glasses because they will leave residue. Use low lint cloths or lens paper to clean glass plates.
 - DO NOT USE CANNED DUST REMOVER TO CLEAN THE SCANNER. THIS CAN DAMAGE THE SCANNER BY LEAVING A RESIDUE ON SENSITIVE PARTS.
 - Negatives may be cleaned using a photographic brush or canned air.

- 2) Load electron micrograph(s) emulsion side down between glass plates.
 - Please note: the glass plates should never touch each other – there should always be a negative or the Zeiss plastic insert between the plates.
 - WEAR GLOVES!
 - Be extremely careful lifting the upper glass plate. Rest the plate on the two spongy pads that are on the underside of the raised scanner lid. The spongy pads do not grip the glass plate very tightly, therefore make sure the upper glass plate is securely held up before letting go.
 - Place Zeiss plastic insert in manilla folder on shelf near scanner. Keep this insert dust-free.
 - Once negatives are in place, slowly lower upper glass plate. Again, be very careful. The glass plate fits snugly in the two, black metal corner guides. Make sure to lower slowly and carefully so the glass is not chipped.

How to launch the scanning software:

- 1) Make a new folder for yourself in one of two places:
Data1 (E:) or Data2 (F:)
E and F hold about 70 GB each.
A 14 micron scan of approximately the entire negative at 8 bits/greyscale is about 40 MB.
DO NOT write anything into Local Disk (C:) or New Volume (D:). That space is needed by the program.
- 2) Launch “Controller Hyper Terminal” (icon on desk top)
This provides the error messages during program runs. You can minimize this window.
Anytime you encounter errors during scanning write down or save the text output from the controller hyper terminal. **Write down the errors in the log book and report the error to Paul.**
- 3) Launch “PhotoScan” (icon on desk top)
“PhotoScan” window appears

How to load configurations:

- 1) Under “File” menu, select “New Scan”
“Scan Properties” window appears
you should see three tabs: scan setup, film/output, camera definition
- 2) Under scan setup, select a parameters file from available list: for example, “cryoEM.par”
Check settings for Scan Mode, should be “Gray Scale” “8 bit”
Adjust resolution (7, 14, etc etc to 224 microns; use 224 for low resolution scan)
Tile size (256)
Format (tiff uncompressed)
Overview (full set)
At bottom of Scan Properties – Scan Setup window, type in name of image file,
making sure to write into your own folder on either E: or F: disks.
- 3) Under Film/Output setup, make sure the parameter file listed matches the one you’re using (ie: cryoEM.par).
Select Film Polarity (negative)
Output polarity (positive)
Output data proportional to: Scan Gamma Correction 100
- 4) Under Camera Definition tab, make sure parameter file name matches, check resolution
Film emulsion should be at “Right Reading”

(The commands in this “how to” guide are provided as options from pull-down menus. There are also icons corresponding to many/most of the commands. To identify the appropriate icon, position the mouse cursor over the icon list – the command to which it corresponds will appear after 1-2 seconds.)

How to “prescan” and make your final scan:

- 1) To make a “prescan” type image, select a low resolution (ie: 224 micron) scan size and adjust the stretchy box (defining a yellow rectangle using left mouse button) so it is likely to cover the entire negative
- 2) Under “scan menu” of PhotoScan window, select scan to disk
A scan status window comes up to show you the progress
After the scan is finished, you should be able to adjust the view area by “Zoom” or “fit images” option under View menu.
- 3) Under “define scan area” menu option (under “View”), pick a smaller rubber banded box of the part of the micrograph you want.
Do not include very dark or very light areas of the negative or surrounding region – it will screw up the histogram.

- 4) Under scan Properties window, go to “histogram tab”. Select a generous portion of the histogram (using the mouse, pick the peak and go **well** past it on either side).
The image is adjusted to account for the new range. Hit the “apply” the histogram button. Note: if you cut the histogram too tightly, you will not be able to adjust the contrast/range of the micrograph properly later on. The “reset” button can be used to undo the change at this step.
- 5) Change resolution (ie: to 7 or 14 microns) in Scan Mode settings page.
- 6) Scan to disk.
It will ask if you want to keep the last scan. Say no (until you achieve your final scan). A scan status window appears during the scan.
It takes about one minute to scan a negative at 14 microns resolution.
- 7) If you like the image, you’re done. Quit and save or go to make a new scan and save the old one when prompted.
- 8) If you don’t like the image, adjust parameters (particularly histogram) until you achieve a good scan.

After you are finished scanning:

- 1) Exit both programs “Controller Hyper Terminal” and “PhotoScan”
- 2) Turn off scanner
- 3) SFTP files over to your machine. (Icon for SFTP program is on desktop.)
- 4) Delete files from the scanner’s computer within 7 working days. Files left on longer are subject to deletion.
- 5) Shut down computer (“shut down” option of Windows).

Scanner Users,

A sign-up system will be implemented for use on the new scanner. Individuals wishing to reserve the scanner can do so by filling in their name and PI in the weekly calendar which is located beside the scanner, in the black binder.

Two-hour time slots have been created for use during peak use periods (8am-6pm, Monday-Friday). Each user may reserve two peak period time slots at a time. Once one of the slots has passed, the person may again reserve time. Thus, if no one reserves the slot immediately following your scheduled time, you may roll your reservation over to the next slot once the first time period has elapsed.

Off hour times from 6pm until midnight and from midnight until 8am, as well as 2 hour weekend 8am-6pm slots are also available. Each user can have two reservations in the off hour time blocks at any given time. These reservations also can be rescheduled as each time period passes.

This is an initial scheduling procedure. If you have problems or ideas on scheduling, you likely have too much free time but can contact Paul Chipman (Lilly Room B-216) for further discussion.

Thanks,

Paul Chipman